FEBS Journal 2010 vol.277 N1, pages 186-196

Binase cleaves cellular noncoding RNAs and affects coding mRNAs

Mitkevich V., Tchurikov N., Zelenikhin P., Petrushanko I., Makarov A., Ilinskaya O.
Kazan Federal University, 420008, Kremlevskaya 18, Kazan, Russia

Abstract

Bacterial RNases are promising tools for the development of anticancer drugs. Neoplastic transformation leads to enhanced accumulation of rRNA and tRNA, and altered expression of regulatory noncoding RNAs. Cleavage of RNA in cancer cells is the main reason for the cytotoxic effects of exogenic RNases. We have shown that binase, a cytotoxic ribonuclease from Bacillus intermedius, affects the total amount of intracellular RNA and the expression of proapoptotic and antiapoptotic mRNAs. For four cell lines, we visualized cellular RNA by fluorescence microscopy, and determined RNA levels, viability and apoptosis by flow cytometry. We found that the level of cellular RNA was decreased in cells that were sensitive to the cytotoxic effects of binase. The RNA level was lowered by 44% in HEK cells transfected with the hSK4 gene of the Ca 2+-activated potassium channels (HEKhSK4) and by 20% in kit-transformed myeloid progenitor FDC-P1iR1171 cells. The most significant decrease in RNA levels was registered in the subpopulations of apoptotic cells. However, the binase-induced RNA decrease did not correlate with apoptosis. Kit-transformed cells with binase-induced RNA decrease retained viability if the interleukin-dependent proliferation pathway was activated. Using quantitative RT-PCR with RNA samples isolated from the binase-treated HEKhSK4 cells, we found that the amount of mRNA of the antiapoptotic bcl-2 gene in vivo was reduced about two-fold. In contrast, expression of the proapoptotic genes p53 and hSK4 was increased 1.5-fold and 4.3-fold, respectively. These results show that binase is a regulator of RNA-dependent processes of cell proliferation and apoptosis. © 2009 FEBS.

http://dx.doi.org/10.1111/j.1742-4658.2009.07471.x

Keywords

Apoptosis, Cellular RNA degradation, Cytotoxicity, Gene expression, RNase